

Experiment #5: Qualitative Absorption Spectroscopy

One of the most important areas in the field of analytical chemistry is that of spectroscopy. In general terms, spectroscopy deals with the interactions of radiant energy with matter (i.e., atoms, ions, or molecules) in various physical and chemical states. This energy is usually some type of electromagnetic radiation, abbreviated EMR, consisting of electric and magnetic components. In most spectroscopic methods, only interactions of the electric component of the EMR are of interest.

Electromagnetic radiation exhibits a “dual nature”, showing characteristics of both a cyclic wave motion and discrete particles of energy called photons. Wave properties explain such phenomena as reflection, refraction, and optical interference, while particulate properties are required to explain the photoelectric effect and optical emission and absorption processes.

The major cyclic wave motion properties are wavelength (λ) and frequency (ν). The wavelength is the distance between corresponding points on the EMR wave, and frequency refers to the number of complete wave cycles which pass a given point per unit time. Thus, the velocity, c , of the EMR wave is given by the equation: velocity = wavelength \times frequency (or $c = \lambda\nu$). EMR is often classified with respect to the number of different energies or wavelengths in the beam of radiation. Monochromatic EMR has only one energy or wavelength; polychromatic EMR has more than one type of energy or wavelength.

The EMR photon characteristics include intensity (P) and energy (E). Intensity describes the number of photons present in the EMR beam or the number of photons emitted by an energy source per unit time. The energy of a photon is given by the equation: $E = h\nu = hc/\lambda$, where “ h ” is Planck’s constant. Thus, the energy of a photon is controlled by the EMR frequency (which is determined by the energy source). As the EMR frequency increases (or the corresponding wavelength decreases), the photon energy increases.

In matter–EMR interactions, radiation may be transmitted (pass through the matter with no measurable effect), absorbed (part or all of the energy is permanently transferred to the matter), or scattered/reflected (the EMR beam direction changes, but no energy is lost). In most chemical systems, absorption is the transition of primary importance. With respect to matter, absorption is viewed as an irreversible, two-step process: (1) energy gain (via transfer of photon energy to the absorbing species, promoting change to a higher energy state followed by (2) energy loss (in which the excited absorber gives up the gained energy via thermal/collisional decay, photochemical reaction, or a photoluminescence process).

Absorption spectroscopy involves interactions over the entire spectrum, ranging from very high energy gamma-rays (which cause nuclear changes) to very low energy radiowaves (which cause spin alignment changes in the atom). Transitions may involve nuclear, electronic, vibrational, rotational, and spin alignment changes, listed in order of decreasing relative energies. In all absorption processes, however, the EMR photon can be absorbed in the chemical species. That is, absorption is an “all-or-nothing” process.

In the absorption of visible light, the transition involves promoting an outer (valence) shell electron from a low energy or ground state orbital into a higher or excited energy state. When a solution illuminated by white light appears colored, some of the light is absorbed, and the color seen is due to the residue. Such a solution may be described in terms of the colors or the EMR wavelengths which

pass through the solution. Thus, a “blue” solution is one which transmits wavelengths corresponding to the color “blue”. However, it is equally valid (and of more interest chemically) to describe a solution in terms of the colors or wavelengths of light which are absorbed. A “blue” solution then, is one which absorbs the color complementary to blue, namely yellow.

Physiologically, the human eye can detect three primary bands of color: magenta, cyan, and yellow. All perceived colors are various combinations of intensities of light in these three ranges! The absorption of light by three different dye-like compounds in the human eye is the first step in the biochemical process of color vision.

A range of instruments have been developed for spectroscopic applications. The fundamental designs are similar, differing only in the specific components used to perform various functions. Instruments are designated as photometers, spectrometers, or spectrophotometers, depending upon the components utilized in the system. Irrespective of the purpose or application, spectroscopic instruments contain similar components, including:

an energy source (to emit stable, continuous EMR of the appropriate energy), a wavelength-control (to select appropriate wavelengths from the source output), a sample cell (to hold the gas, liquid, or solid absorbing species in the EMR path), a detector (to measure incident and emergent EMR intensities), and a recorder (to give a mechanical or electrical response to the detector output).

Instruments which use only visible light are called “colorimeters”, and the Spectronic 20 is a typical member of this group. The source consists of an incandescent-lamp with a tungsten filament which glows when heated; the cell is a glass cylinder; the wavelength-control system is made up of slits, lenses, and a reflection grating; the detector is a phototube containing a cathode coated with a photosensitive alkali oxide which emits electrons when struck by photons and an anode which collects the released electrons to give a complete flow of electricity; and a meter which converts the phototube current into a digital read-out.

The Spectronic 20 source emits EMR wavelengths over the entire visible region, but not in equal intensities. In fact, the source emits much more red and infrared radiation, while giving off much less violet and ultraviolet radiation.

Conversely, although the detector responds to all visible wavelengths, it is more sensitive toward the more energetic ultraviolet end of the EMR spectrum. This opposing interplay of source intensity and detector sensitivity must be balanced at each different wavelength of interest.

The Spectronic 20 wavelength-control system is rather simple in design and capability. The reflection grating contains 600 grooves/mm and provides a wavelength accuracy of ± 2.5 nm, although the meter scale has a wavelength readability of 1 nm. The grating dispersion is of minor quality, resulting in a nominal spectral width of 20 nm across the entire range from 340 nm to 950 nm. Thus, the instrument can completely separate and differentiate two wavelengths only if they differ by more than 20 nm.

The Spectronic 20's will generally be turned on and warmed up when the students arrive in the laboratory. If not, the turn-on is as follows: Using the left control, turn the Spectronic 20 on, and allow a five minute warm-up period.

EMR wavelength and visual color correlations

Each student will make a correlation series of wavelengths and perceived colors, using the EMR spectral output of a Spectronic 20 colorimeter. Place a slanted piece of white chalk in a clean, dry

cuvette, and insert the cuvette into the sample compartment. Leave the compartment cover up so the EMR beam can be observed as a reflection on the smooth, slanted surface of the chalk. Leave the left control in the counter-clockwise position, turned just enough to turn on the instrument. Looking down at the chalk, turn the right control until the color band can be clearly seen.

Set the top wavelength dial to 400 nm, and examine the color of the light band shown on the chalk. Record a personal description of the observed color in Table I. Change the wavelength dial to a new setting (using 25 nm intervals) and repeat the process. It may be necessary to change the position of the right control to sharpen the color band as the wavelength is changed. Record the associated color at each wavelength setting from 400 nm to 625 nm.

Bausch & Lomb Spectronic 20^R



Table I

λ , nm	Color Description

Dye absorption spectrum

Select a dye solution from the reagent shelf. If working in groups, your instructor will instruct you as to whether each student should select a different dye solution, or whether the same dye solution will be used by each member of the group. Record the name of the dye (if it is provided) and the visible color exhibited by the solution. Fill a clean cuvette half full with the dye solution, and fill a second cuvette with deionized water to use as a blank or reference solution.

With the Spectronic 20 cell compartment empty and closed (and the instrument turned on and, warmed up), set the wavelength dial to the desired starting wavelength (i.e., 400 nm). Adjust the left control to place the meter needle or digital display exactly on 0% T. Place the blank solution in the cell compartment, close the cover and adjust the right control to set the needle or digital display to 100% T. Each time the wavelength is changed, this standardization procedure must be carried out.

Replace the blank solution with the dye solution, close the cover, and observe the needle position on the meter scale or digital readout. Record the experimental percent transmittance and absorbance, making sure to read the scales correctly. Since the transmittance scale is linear and the absorbance scale is exponential, it is better practice to record both experimental %T and A values, and then use the %T value to calculate a more precise value for absorbance, using the equation: $A = 2 - \log \%T$.

Complete Table II entries at each wavelength from 400 nm to 625 nm, using wavelength increments of 25nm to cover the spectral range. Remember to zero the instrument with the blank solution at each new wavelength. Allow each member of the group to read and record experimental % T and A values for his or her indicator solution before changing the wavelength.

Table II

λ , nm	Experimental A	Experimental %T	Calculated A

Data treatment and report

A three-page report must be submitted by each student. Each page must contain a complete heading, listing: experiment title, student name, names of members in the group, instructor's name, class number and section number, and date on which the report was submitted.

The first page of the report will contain a neat, complete titled, copy of the data given in Table I. All information taken in the laboratory procedure must be included, using a format identical to that of Table I.

The second page of the report will consist of a titled copy of Table II, giving all data recorded in lab. In addition, the specific name of the dye must be reported if known. The information should be examined determine the light colors and wavelengths which are best transmitted by the dye solution as well as the colors and wavelengths which are best absorbed. Conclusions about the regions of transmittance and absorption should be expressed in a summary statement.

The third and final page of the report will consist of the absorption and transmittance spectra of the indicator solution. On the same sheet of graph paper (18 x 24 cm), make two separate plots, one of % T vs. λ and a second of calculated A vs. λ . The following specifications and instructions must be obeyed in performing the graphical data treatment.

The long axis must be used as the abscissa (horizontal axis), upon which the wavelength (in nm's) is scaled in increasing values from left to right. The left ordinate (vertical axis) is to be used for the calculated absorbance values, while the right ordinate (vertical axis) will be the % T axis.

Use independent linear scales for each axis, and maximize the scale values to use most of the graph paper. The % T axis should be scaled to go from a starting value of 0% T to an upper value of 100% T. The A axis should begin at 0, but the upper value should be chosen to expand the data points over most of the graph sheet.

Connect the data points of each curve in a smooth, continuous line. Do not make connect-a-dot curves. Include a plot title and complete heading on the graph page, and make sure to identify the specific dye used in the spectroscopic measurement.